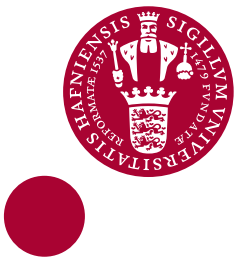

Broad spectrum antimicrobial activity of Bacillus amyloliquefaciens strains isolated from traditional Bikalga, an African fermented food condiment

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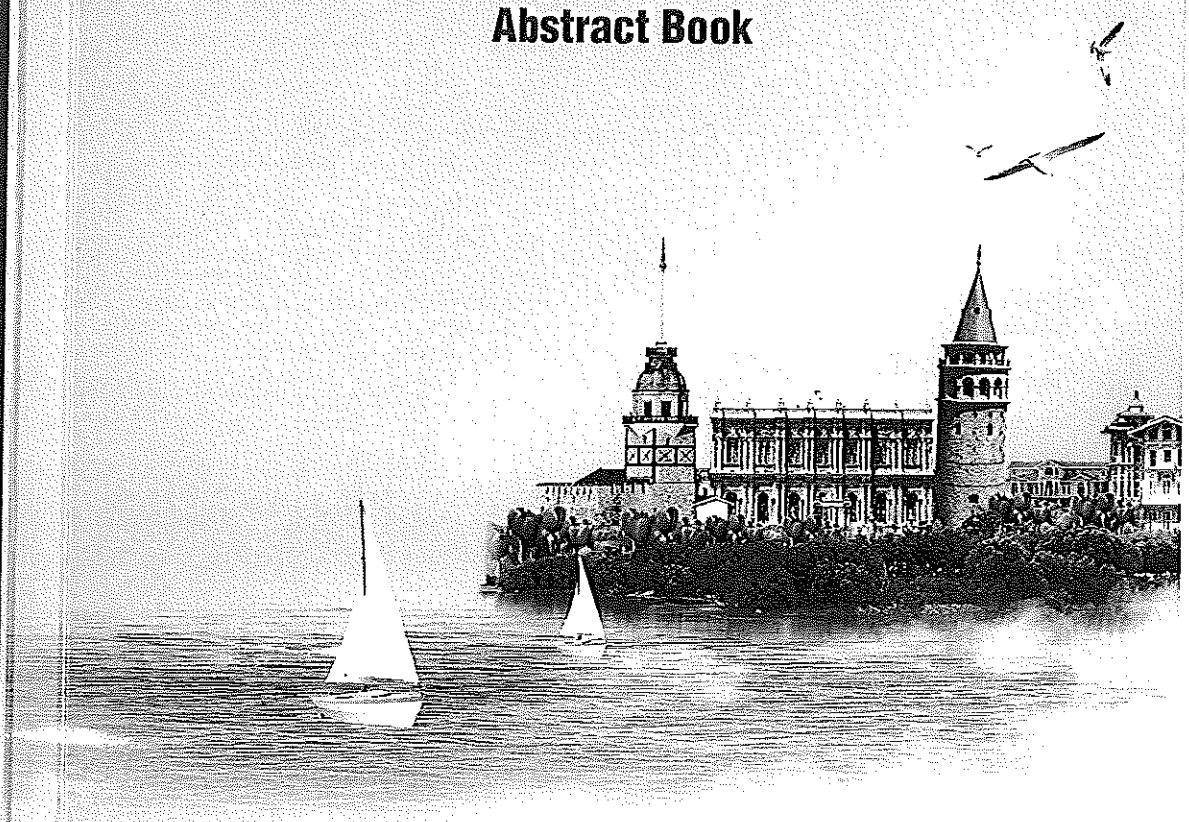

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Broad spectrum antimicrobial activity of *Bacillus amyloliquefaciens* strains isolated from traditional Bikalga, an African fermented food condiment

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Bikalga is a West African food condiment made from spontaneously fermented seeds of the plant Hibiscus sabdariffa. The fermentation is alkaline and dominated by Bacillus species. Three *Bacillus amyloliquefaciens* isolates (A4, I8, G3) belonging to the predominant aerobic sporeformers of bikalga were screened against a panel of 40 indicator organisms for their antimicrobial activity using agar spot and agar well diffusion assays. Antimicrobial activity in BHI medium was observed against several *Penicillium* spp. as well as a wide range of Gram-positive and Gram-negative food borne pathogenic bacteria, including *Bacillus cereus*, *Listeria monocytogenes*, *Salmonella* spp., *Escherichia coli*, *Shigella dysenteria* and *Yersinia enterocolitica*. The investigated isolates further produced antimicrobial activity against *Bacillus cereus* in a Hibiscus sabdariffa seeds based medium. The antibacterial activity against *B. cereus* in BHI-broth was detected from the middle of the exponential phase, and maximum activity was observed during stationary phase. Treatment of cell free supernatants (CFS) to temperatures of 4°C and up to 80 °C, pH 3-11, and proteolytic, amylolytic and lipolytic enzymes did not affect the antimicrobial activity against *B. cereus*. Different extraction methods were used to obtain and further characterize the antimicrobial substance produced. No antimicrobial activity was observed for proteins > 2.5 kDa (precipitated from CFS using tricloroacetic acid) as deduced from SDS page gels overlaid with *B. cereus*. In comparison ethyl-acetate extracts of CFS showed antimicrobial activity against both the Gram-positive and Gram-negative indicator bacteria. HPLC-HRMS analysis of ethyl-acetate extracts with antimicrobial activity revealed the presence of iturins in addition to other compounds. Iturins are cyclic lipopeptides produced by various *Bacillus* spp. found to mainly inhibit yeast and fungi. Our results suggest that the three investigated *B. amyloliquefaciens* isolates produce more than one compound with antimicrobial activity. The antimicrobial activity against the Gram-positive and Gram-negative indicator bacteria might be associated to production of small lipophilic compounds other than iturins. In conclusion the *Bacillus amyloliquefaciens* strains of Bikalga show some interesting antimicrobial properties. They may have potential for use as starter cultures for the biological control of pathogenic and spoilage microorganisms during bikalga fermentation, though further investigations are needed.

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In situ production P572 in cheese

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Reuterin (β-hydroxypropiona by some strains of *Lactoba* positive and Gram-negative *in situ* production of reuterin considered an alternative for isolates of *L. reuteri* (INIA P5 P577 and INIA P579) that sh gel electrophoresis (PFGE) g Reuterin concentrations obt supplemented with 100 mM o Cheese was manufactured concentrations of 50, 100, 2(commercial starter culture an control cheese was made from was observed during the 3(supplemented with glycerol b from the β-HPA. The highe detected on day 1 in cheese concentrations corresponded indicator microorganism. In co after 30 d, while the addition reductions of 5.33 log units o glycerol, counts of 6.09 log cfu reduction of *L. reuteri*, with va to levels in control cheese. Val ripening period. According to o the development of a bioprotect